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Does environment affect polymorphism of B chromosomes in the yellow-necked mouse *Apodemus flavicollis*?

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B chromosomes are chromosomes additional to the standard complement. Their only consistent feature is that they are not essential for survival of an individual. Contrary to their widespread occurrence in plants and insects, they have been found so far in only 35 species of mammals (Vujošević 1993). B chromosomes are a widespread component of the genetic system of the yellow-necked mouse *Apodemus flavicollis*. In almost all the studied populations animals with Bs were present (Soldatović et al. 1975; Kral et al. 1979; Sablina et al. 1985; Giagia et al. 1985; Vujošević et al. 1991; Nadjafova et al. 1993; Zima and Macholán 1995; Boeskorov et al. 1995) with frequencies varying from 0.11 in Slovenia to 0.94 in the Western part of the Czech Republic.

B chromosomes in *A. flavicollis* are acrocentrics as are the chromosomes of the standard complement. Equilibrium frequencies of Bs were found in the course of year-to-year studies regardless of changes in population density (Vujošević 1992). On the other hand, it was found that seasonal changes in frequency of animals with Bs could be significant under conditions of stress produced by overcrowding (Blagojević and Vujošević 1995) or could follow changes in population density if abundance is moderate and there is no increased competition for food and space (Vujošević and Blagojević 1995). A number of studies in different taxonomical categories has shown that the presence of B chromosomes in certain populations can be associated with certain ecological and climatic variables (Fröst 1958; Bosemark 1956; Brown 1960; Parker et al. 1991, Confalonieri 1995). As Bs are found in different regions throughout the range of *A. flavicollis* at different frequencies, it was of interest to examine whether or not the environment influences their appearance in any way. In this study the status of B chromosomes in Yugoslavian populations of *A. flavicollis* is investigaied concerning correlations to certain environmental variables.

A total of 967 live specimens of the yellow-necked mouse *Apodemus flavicollis* was collected using "Longworth" traps at following localities in Yugoslavia (coordinates of the UTM system are given in brackets): Ada (CQ82), Avala mt. (DQ64), Cer mt. (CQ84), Donji Dobrić (CQ83), Goč mt. (DP82), Fruška Gora mt. (DR00), Jastrebac mt. (EP30), Petnica (DQ10), Maljen mt. (DP28), Lešnica (CQ82), Kopaonik mt. (DN88) and Rudnik mt. (DP68). In this number animals previously studied are also included (Vujošević and Blagojević 1995; Blagojević and Vujošević 1995). Chromosome preparation was done directly from bone marrow cells using the standard technique (Hsu and Patton 1969). G- and C-banding were performed using the slightly modified procedures of Seabright (1971) and Summner (1972). Climatological data (average temperature, average number of tropical days, average number of sub-zero temperature days, rainfall, and snowfall) were obtained from the Federal Hydrometerological Institute and represent

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Table 1. Environmental data for the studied localities (Alt – altitude, AT – average temperature, ANTD – average number of tropical days, ANSD – average number of sub-zero temperature days, RF – rainfall, SF – snowfall

	Locality	Alt. (m)	AT (°C)	ANTD	ANSD	RF (ml/m²)	SF (cm)
1	ADA	150	11.0	40	25	750	30
2	AVALA	450	10.0	10	35	750	30
4	D. DOBRIĆ	300	11.0	25	15	850	30
5	GOČ	750	7.0	10	60	950	55
6	F. GORA	400	10.5	30	30	750	30
7	JASTERBAC	500	7.0	5	45	850	55
8	PETNICA	250	11.0	40	25	750	30
9	MALJEN	900	7.0	10	50	950	70
10	LEŠNICA	150	11.0	40	25	750	30
11	KOPAONIK	1 700	3.0	0	85	850	130
12	RUDNIK	750	7.5	10	50	950	70

Table 2. Results of analysis for the presence of B chromosomes at different localities (fB – frequency of animals with Bs; fB/B – frequency of Bs per B carrying animal). (*) data from Blagojević and Vujošević (1995); (**) data from Vujošević and Blagojević (1995).

	Locality	N	0 B	1 B	2 B	3 B	4 B	5 B	fB	fB/B
1	ADA	64	52	9	3				0.19	1.25
2	AVALA	32	19	8	4	1			0.41	1.46
3	CER (**)	132	88	30	12	2			0.33	1.36
4	D. DOBRIĆ	43	33	8	1	1			0.23	1.30
5	GOČ	19	10	6	2	1			0.47	1.44
6	F. GORA	29	18	7	1	2	1		0.38	1.73
7	JASTERBAC (*)	530	337	136	47	5	4	1	0.36	1.38
8	PETNICA	19	12	4	1	2			0.37	1.71
9	MALJEN	20	11	4	4	1			0.45	1.67
10	LEŠNICA	44	31	9	4				0.29	1.31
11	KOPAONIK	24	9	13	2				0.63	1.31
12	RUDNIK	11	4	5	2				0.62	1.29
	TOTAL	967	624	239	83	15	5	1	0.35	1.38

average data for fifty years (Tab. 1). Frequencies of B chromosomes were transformed according to the method of Christiansen et al. (1976) which takes into account differences in sample size. The regression analyses was carried out using the software STATISTICA for Windows (StatSoft, Inc. 1995).

B chromosomes were present in all the samples studied with an average of 0.35 and ranging from 0.19 to 0.64 (Tab. 2). Due to the presence of animals with different numbers of Bs, the frequency of Bs per B carrying animals varied from 1.13 to 1.73 in the different populations. Among animals with Bs the most frequent were those with one B chromosome (0.70). Two Bs per animal were found in all populations making a frequency of 0.24 in the sample. Animals with three Bs were absent from four populations, while four Bs were found in two populations only. Only one animal with 5 Bs was found. There was no significant difference between the frequency of Bs in males and females.

The frequency of animals with Bs in the different populations correlated significantly with the environmental variables $R = 0.94$ ($F_{(6,5)} = 6.14$; $p < 0.03$). The average temperature ($r = -0.79$; $p < 0.005$) and the average number of sub-zero temperature days ($r = 0.82$;

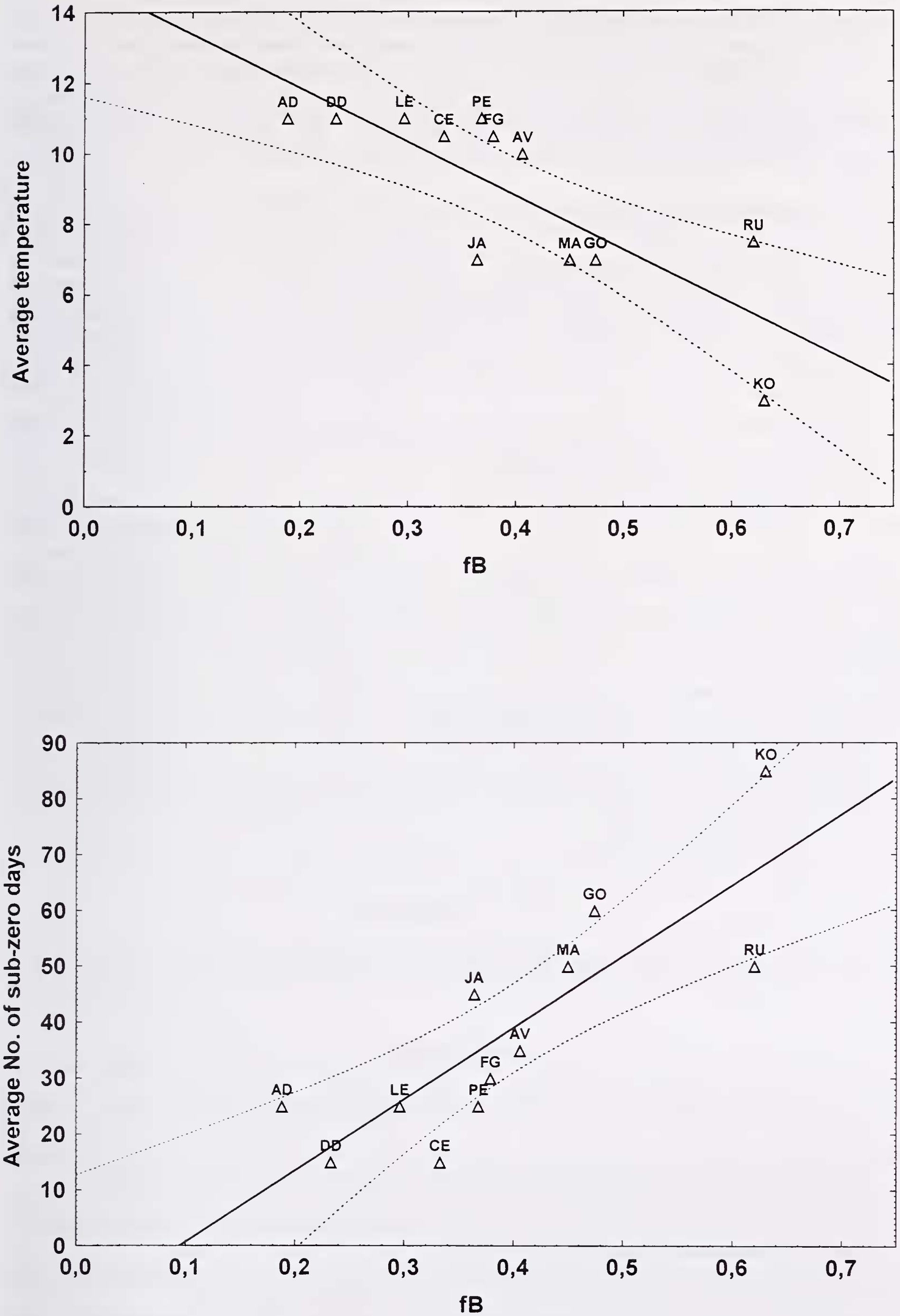

Fig. 1. Correlation between frequency of animals with Bs (fB) and average annual temperature (upper graph) and between frequency of animals with Bs (fB) and average number of sub-zero temperature days (graph below). Populations are labeled with acronyms.

$p < 0.002$) significantly influenced B frequencies. The frequency of B-carriers in populations was negatively associated with the average temperature and positively with the average number of sub-zero temperature days (Fig. 1). Temperature undoubtedly represents the most critical climatic factor and it could be said that *A. flavicollis* with Bs do best at extreme temperatures. As altitude is mainly a measure of the temperature and exposure it is not surprising that with increasing altitude the frequencies of animals with Bs also increases.

Only a small proportion of species possessing Bs has been surveyed extensively. Results of the survey of different plant species (Fröst 1958; Bosemark 1956; Brown 1960; Parker et al. 1991) led to the conclusion that B chromosomes reached the highest frequency under conditions that were most favorable for growth of the species concerned. Barker (1966) and Hewitt and John (1967, 1970) who showed the existence of a negative correlation between B chromosome frequency and rainfall obtained similar results for the grasshopper *Myrmeleotettix maculatus*. Confalonieri (1995) found in an analyses of 25 samples of the grasshopper *Trimerotropis pallidipennis* from Argentina that the frequency of B-carriers in populations was associated with altitude (negatively) and longitude (positively). The author claimed that the higher incidence of B chromosomes was associated with more favorable environments.

Our results show the opposite trend. Frequencies of animals with Bs are highest in extreme climatological conditions. Holmes and Bougourd (1989) described in *Allium schoenoprasum* a situation where B chromosomes conferred a selective advantage to the survival of individuals during the early stages of the life-cycle. Higher frequencies of animals with Bs in peripheral populations were found in the harvest mouse *Reithrodontomys megalotis* (Shellhammer 1969) and pocket mouse *Perognathus baileyi* (Patton 1972). As the most likely explanation Shellhammer (1969) mentioned the general increase in genetic variability towards the periphery of the distribution area of a species. It could be added that high altitude produces similar effects on the genetic structure of species as does the periphery of its distribution. Therefore it is possible that in *A. flavicollis* the presence of Bs increases the ability of the species to occupy such an area. It is difficult to decide whether increased levels of B's are a consequence of stress or whether they provide better resistance against stress.

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